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TECHNOLOGISTS OF SOUTH AFRICA

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September, 1961



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vir
Mediese Laboratorium
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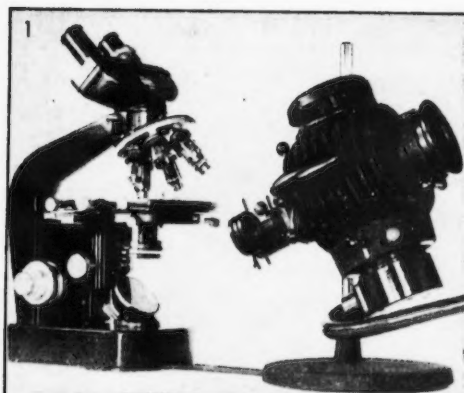
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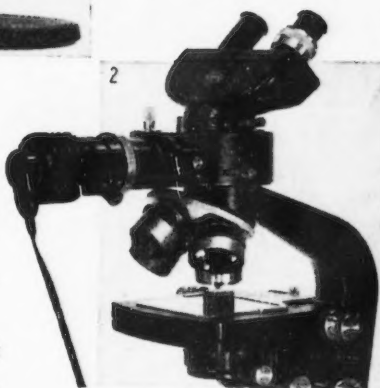
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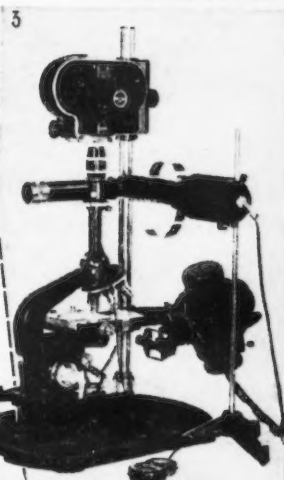


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*Current Affairs**The Editor*

THE INTERMEDIATE EXAMINATION

THE COURSE OF INSTRUCTION leading to the Intermediate Examination is under fire at the moment and although maximum heat appears to be generated at the tip of the continent there are wider spread foci of dissatisfaction.

It appears that the present syllabus is criticised because it contains "too much Clinical Pathology", and is accused of bias in favour of the candidate who will subsequently take "Clinical Pathology" as a final subject. We may perhaps be forgiven if we say that we entirely fail to see the logic of this accusation. Surely the purpose for which the Intermediate Examination was designed was to ensure that no student went on to the study of a specialised subject, such as Virology or Blood Transfusion Technique, who was not adequately grounded in general routine laboratory work? If the amount of general laboratory work, such as the estimations of blood sugar, urea etc., is to be reduced are we not, in the future, going to be faced with a race of hyper-specialists who, whilst first-class workers at one subject, cannot be used to relieve congestion in some other part of the laboratory? In academic spheres and in closed public health and hospital laboratories this may not seem to be an insurmountable problem, but to the organisers of widespread laboratory complexes it is a devastatingly real problem. It is often necessary in emergency to switch staff amongst constituent laboratories of such a service and if all the available staff happened to be "single subject specialists" chaos would result.

A further point is that a broader basic laboratory education produces a better understanding of laboratory medicine as an entity.

We appeal to all concerned not to be too hasty or too parochial in their approach to this problem.

*Huidige Gebeure**Die Redakteur*

DIE INTERMEDIËRE EKSAMEN

DIE OPLEIDINGSKURSUS in voorbereiding vir die Intermediêre Eksamen kom tans onder skoot en, alhoewel die Suidpunt van die vasteland die mees ontstoke voel oor dié saak, is daar nietemin ander kolle deur die land waar ontevredenheid heers.

Skynbaar word die huidige sillabus daarvan beskuldig dat dit „te veel kliniese patologie” behels en dat dit bevoordeel is ten gunste van die student wat later kliniese patologie as ’n eindvak sal hê. Ons hoop dit sal ons nie ten kwade gereken word as ons nie die logika van hierdie aantyging kan raaksien nie. Die oogmerk waarmee die Intermediêre Eksamen opgestel was, was sekerlik dat geen student veronderstel word om te studeer in enige gespesialiseerde vak, soos virologie of bloedtransfusietegniek, sonder dat hy eers die grondbeginsels van algemene roetine laboratoriumwerk deeglik onder die knie gekry het nie. As die hoeveelheid algemene laboratoriumwerk soos bv. bloedsuiker en ureembepalings verminder word, loop ons dan nie die gevaar om in die toekoms ’n klomp „hiper”-spesialiste te hê wat, hoewel hulle eersterangse werkers op ’n besondere gebied mag wees, tog nie gebruik kan word om druk in ’n ander gedeelte van die laboratorium te kan help verlig nie?

Dit mag alles goed en wel wees in akademiese inrigtings, in sommige „geslote” publieke gesondheids- en hospitaallaboratoria, maar waar wydverspreide laboratoriumdienste bestaan, is dit ’n ware probleem en een wat baie hoofbrekens gee. Dit is dikwels nodig om in noodgevalle staflede uit te ruil tussen laboratoria en, sou hulle nou almal „enkelvak spesialiste” wees, sal wanorde sekerlik aan die orde van die dag wees.

Ten gunste van ’n breër opleiding in die basiese laboratorium-tegnologie, moet ’n mens ook in ag neem dat daar ’n beter begrip opgewek word van algemene laboratoiumgeneeskunde as ’n entiteit op sy eie.

Ons versoek is aan alger om nie te haastig of te eng in hulle benaderinge van hierdie probleem te wees nie.

BLOOD GROUP C

by F. A. WARD and V. F. HIGGS

Natal Blood Transfusion Service

ALTHOUGH the vast majority of the specimens which reach a blood grouping laboratory are readily grouped in regard to the ABO system, occasionally specimens arrive which give rise to difficulty. It is, in fact, the occurrence of these oddly reacting cells which partly justifies the existence of blood grouping laboratories as separate entities. Everyone can do a straight-forward ABO group, but not everyone can deal with the difficulties which arise in some cases.

Recently a specimen of blood was received at this laboratory. It was from a Bantu woman nearing the end of her pregnancy. On first testing this specimen, the following reaction was obtained.

Anti-A	anti-B	O serum	A cells	B cells
—	—	2+	4+	4+

This, therefore, looked like a weak group A with a strong anti-A in the serum. The serum was then mixed with A₂ cells and the most unexpected result occurred. There was a 2+ agglutination reaction. The serum was then mixed with two other specimens of group A₂ cells and in each mixture there was moderately strong agglutination. All these results were obtained at 4°C, 22°C and at 37°C.

At this stage it was thought that the antibody agglutinating the A cells might be of a specificity other than anti-A. The serum was therefore tested with cells of seven members of a group O panel. There was no reaction detected by the saline, indirect Coombs, or bromelin techniques.

All these results were confirmed on a subsequent specimen obtained from the patient and in addition a weakly reacting cold antibody was detected.

Opinion at this stage was divided on the question of whether this was a weak group A or a group O. The cells were accordingly mixed with ten potent group O sera, all except one of which agglutinated them. Thinking that the cold antibody was in some way interfering with the reaction of the cells, they were washed three times in saline warmed to 37°C. They were still however agglutinated by group O serum.

By this time the patient had given birth to her baby and on being tested, the baby's cells gave a reaction similar to the mother's. The baby's serum however contained no anti-A, although it did contain a strong anti-B. The cells of two other children of this woman were tested. Of these one was an ordinary group A and the other was a weak group A. Unfortunately the father's blood was not available for testing.

The outstanding attributes of this specimen were:

1. the negative reaction of the cells with anti-A.
2. the positive reaction of the cells with the majority of O sera.
3. the positive reaction of the serum with A cells.

In view of these findings it seems that the most appropriate name for these cells is that suggested by Wiener, namely group C. This must not however, be confused with the "C" of the CDE system of Rhesus nomenclature. According to Wiener's view, the C factor is a factor common to both group A and group B. The existence of this factor is shown by the following observations.

If A cells are injected into an O recipient the recipient may produce a high titre serum not only against A cells but also against B cells. Similarly if B cells are injected into an O recipient the recipient may increase his titre not only against B but also against A. Furthermore if O serum is absorbed with A cells the anti-B activity is reduced as well as the anti-A activity, and the same occurs if O serum is absorbed with B cells. The most reasonable way to interpret these findings according to Wiener is to postulate the presence of a common factor (called C) on both A and B cells, and the presence of not only anti-A and anti-B but also of anti-C in O serum.

O serum therefore, agglutinates A cells not only by virtue of its anti-A content but also by virtue of its anti-C. This specimen therefore is most reasonably called group C, C being the only factor detected on them. The sera of most group C's agglutinate both A cells and B cells, and this, as already indicated, was our finding in the present case.

That the baby's cells gave similar reactions to the mother's need not at the moment concern us as the A agglutinin is often ill-developed at birth. The presence of anti-B and the absence of anti-A however, in the baby's serum, is an interesting finding for which we know of no explanation.

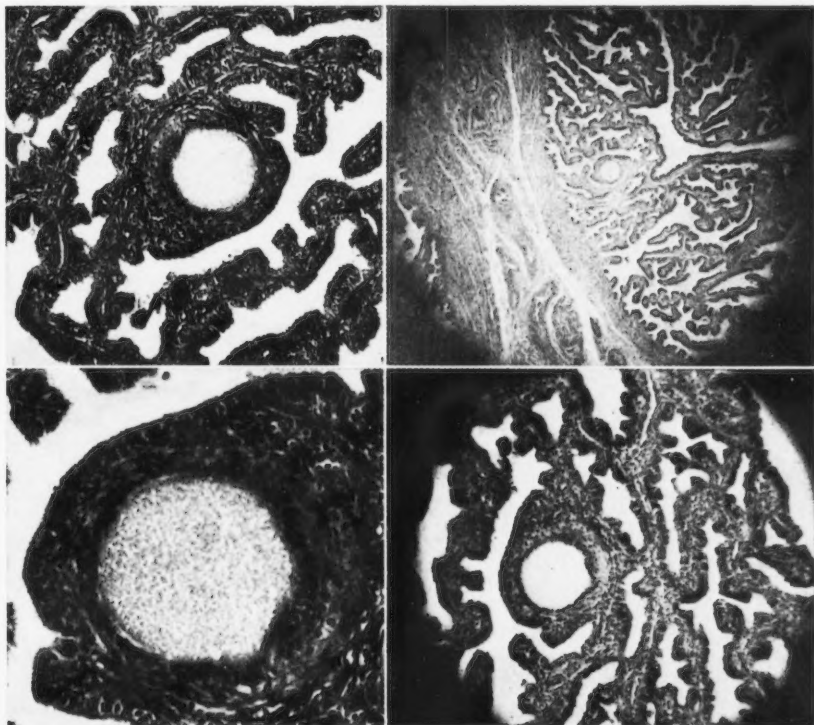
AN EARLY HUMAN OVUM

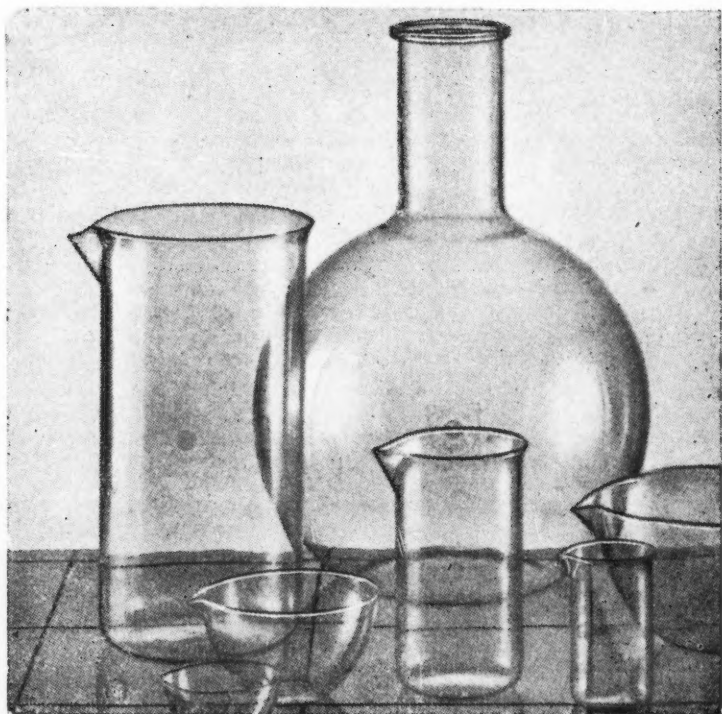
by F. A. WARD

ON THE 23RD JUNE, 1958, a young unmarried woman had an operation performed on her for what was thought to be an ectopic pregnancy. During the operation a fallopian tube was removed which was later examined microscopically. The microscopic appearances at various magnifications, of a section of the tube are shown in the photomicrographs.

In view of these findings the patient was subsequently questioned about sexual intercourse, to which she admitted having on the 7th and again on the 21st June. That is to say, the last act of sexual intercourse occurred two days before the day of the operation, and probably in the region of 40 hours before it.

Whether the object in the fallopian tube is an ovum or not is a question about which there may not be general agreement. I claim that it is an ovum and I leave it to those who disagree with me to prove their point.





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OBSERVATIONS ON THE A AGGLUTINOGEN

by F. A. WARD and V. F. HIGGS
Natal Blood Transfusion Service

NO AGGLUTINOGEN gives rise to more difficulties than A. This is because the A agglutino-gen may occur in various weak forms. These weak varieties of A give difficulty under four circumstances.

1. When they occur alone.
2. When they are associated with B.
3. When they are associated with anti-A.
4. When they are associated with B and anti-A.

Of these, the first is the least, and the last is the most difficult situation.

Although the magnitude of the problem cannot be measured at the present time, it will be apparent from the figures which follow that this is a very common problem among the non-European races of South Africa.

As the anti-A serum in use in this laboratory causes strong agglutination with A₂ cells, these weak variants are not distinguished from A₁ cells. The figures therefore, refer only to those very weak variants which either do not agglutinate or agglutinate very feebly with anti-A. This feeble agglutination is recorded here as a plus one reaction.

Among 4,500 consecutive group A's of non-European origin, the following weak reactions were obtained.

Anti-A	Anti-B	O Serum	
—	—	—	16 times
—	—	+	45 times
1+	—	+	36 times
Total:			97

From these figures it is seen that approximately one out of every 45 group A bloods among non-Europeans possess a weak A agglutino-gen. Furthermore, of these about one in 280 are so weak that they fail to be agglutinated even by group O serum.

When the above figures are broken down into the three racial groups concerned, the following is found.

	Anti-A	Anti-B	O Serum	
<i>Bantu:</i>	—	—	—	13
	—	—	+	42
	1+	—	+	27
	Total:			82 out of 3,000
<i>Indians</i>	—	—	—	1
	—	—	+	Nil
	1+	—	+	4
	Total:			5 out of 1,000

Coloureds	—	—	—	2
	—	—	+	3
	1+	—	+	5
			—	
	Total:			10 out of 500

The specimens which failed to agglutinate with group O serum would have been regarded as group O had it not been for the minor grouping test when the serum either failed to show the expected anti-A activity or showed it only very feebly. Of the 16 specimens mentioned above which failed to react with group O serum, only one showed anti-A activity in the serum and that was only a 1+ agglutination.

It is well known that weak group A's often contain anti-A in their sera and that sometimes this activity is so strong as to produce a plus 4 agglutination with A₁ cells. There is thus every reason to suspect that blood specimens which are believed to be O are in fact weak A's with anti-A. This grouping problem has already been discussed in a previous paper in this Journal where it was recommended that the difficulty be overcome by using A₂ cells in the minor grouping test. The idea behind this suggestion was that the anti-A found in group A's would not agglutinate these A₂ cells.

While more recent experience has justified this recommendation, it must be pointed out that it does not provide the complete answer to the problem. We have in fact encountered a few weak A's, the serum of which agglutinated A₂ cells. One of these was reported in the Journal also.

So although the problem of distinguishing weak A's with anti-A from O has been greatly minimised, it has not been completely solved. The problem of distinguishing weak AB's from B is much greater than that of distinguishing weak A's from O because the use of O serum does not contribute to the solution of the former whereas it contributes very materially to the solution of the latter. It is still therefore, highly probable that not only are a number of A's mis-grouped as O but that a number of AB's are mis-grouped as B.

The following case history illustrates this difficulty and also provides some evidence that such an error is not a serious one. R.N., an adult Indian woman was admitted to hospital with severe burns. She was grouped as B, Rhesus positive. After she had several infusions of plasma, in the course of which she, incidentally, had an allergic reaction, she was given one pint of blood which gave the typical reactions of B, Rhesus positive blood. She received the transfusion well and the nurse in charge noticed no untoward reaction whatsoever until the patient passed black urine.

At this stage, a haemolytic transfusion reaction being suspected, the appropriate specimens were sent to the laboratory for investigation. On re-grouping the donor blood using standard antisera the reactions typical

of B were found. When a highly avid immune anti-A serum was used however, a plus 1 reaction was noted, showing that the donor blood was in fact not B but AB. The urine was tested and while albumin and red blood cells were found there was no trace of free haemoglobin. A biochemist reported that the black pigment had absorption bands characteristic of myoglobin. The level of serum bilirubin remained normal throughout. Within hours the patient's urine cleared and the urinary output was satisfactory at all times.

From this case we see again the capital difficulty of detecting certain A's especially if associated with anti-A and more especially if associated with B in addition. From it we also see that the administration of these very weak AB's to a recipient of group B is not necessarily harmful. Further evidence of this was provided by the observation that the donor blood in the present case was previously given to another group B patient with no harmful results.

It is nevertheless a disturbing thought that certain A's continue to be mis-grouped as O and certain AB's as B, despite the fact that antisera conforming to very high standards are in use.

In an endeavour to find a solution to this problem a weakly reacting group A blood was selected and tested by several techniques and several different batches of anti-A serum.

Centrifuge technique. It is generally agreed that the "spin" group, although not suitable for large scale ABO grouping, is more sensitive than the "standing" technique. Nevertheless only four out of ten anti-A sera caused agglutination and in each of these it was only a one plus reaction.

The use of Bromelin. As bromelin has such a remarkable effect when used with incomplete antibody it was thought that it might enhance the action of complete antibody. In the "standing" technique there was no reaction but when in addition, the tubes were spun, four positive results were recorded two of which were one plus reactions, one a two plus and one a three plus.

The use of Anti-H. An anti-H reagent obtained from the seeds of *Ulex Europaeus*, was diluted in such a way that it gave a faint but definite reaction with O cells. At this dilution however, it caused microscopic agglutination of the weak A cells.

The Reduction of titre method. An anti-A serum was titrated before and immediately after absorption by the test cells. There was no significant reduction in titre.

The Elution method. After absorption, the test cells were washed and the antibody eluted. The eluate caused a plus four reaction with A cells using the spin technique.

Thus it seems that there is no easy way of detecting these weak A agglutinogens. The only technique which gave satisfactory results,

namely the elution method, could certainly not be applied to routine blood grouping work.

We therefore must conclude that a systematic error is being made in blood grouping work for which there is no easy or satisfactory solution. The extent of this error is unknown as is the consequence. There is reason to believe that while the former is large, the latter is not serious.

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2. "Blood Group C", Ward, F. A. and Higgs, V. F. *The S.A. Journal of Medical Laboratory Technology* — this issue

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Hedley J. B. Atkins, Editor. Blackwell Scientific Publications Ltd., 24-25 Broad Street, Oxford. 37s. 6d.

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BOOKS

"Tools of Biological Research" (2nd Series). Edited by Hedley J. B. Atkins

"A Guide to the Identification of the Genera of Bacteria"

V. B. D. Skerman

"Section Cutting in Microscopy." H. F. Steedman.

"Principles and Methods of Clinical Chemistry." F. W. Rice.

CORRECTION

RHESUS INHERITANCE — F. A. Ward. Vol. 7. No. 2. page 24

In paragraph 5 line 4 should read:

Wiener: Anti-Rh₀ anti-rh' anti-rh" anti-hr' anti-hr"

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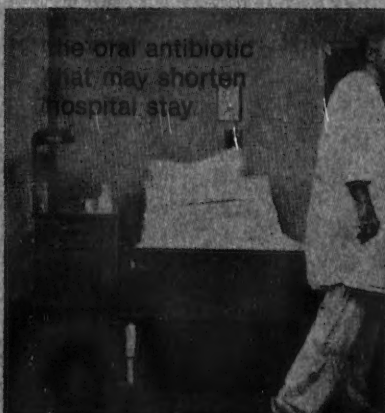
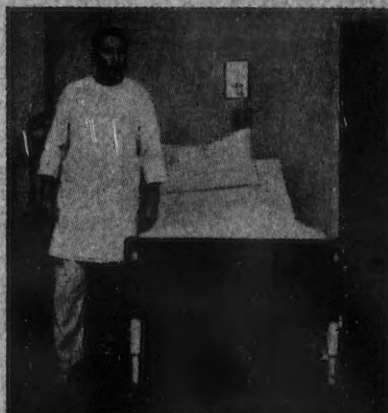
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